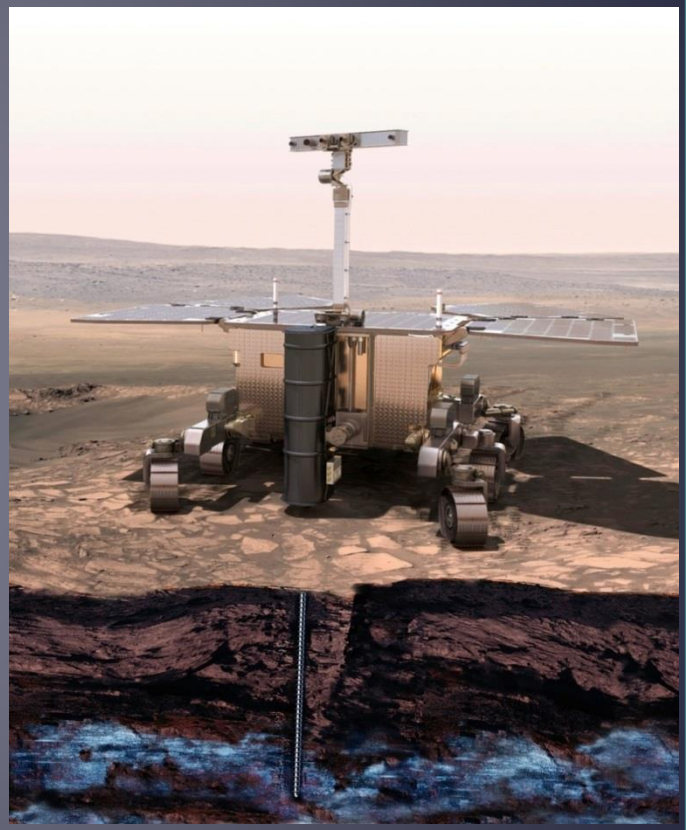
Establishing and Monitoring an Aseptic Workspace for Building the MOMA Mass Spectrometer

Contamination, Coatings, Materials, and Planetary Protection
July 29-31, 2015

Dr. Erin Lalime, SGT/GSFC
Code 546 Contamination and Coatings Engineering Branch

Mars Organic Molecule Analyzer Overview:

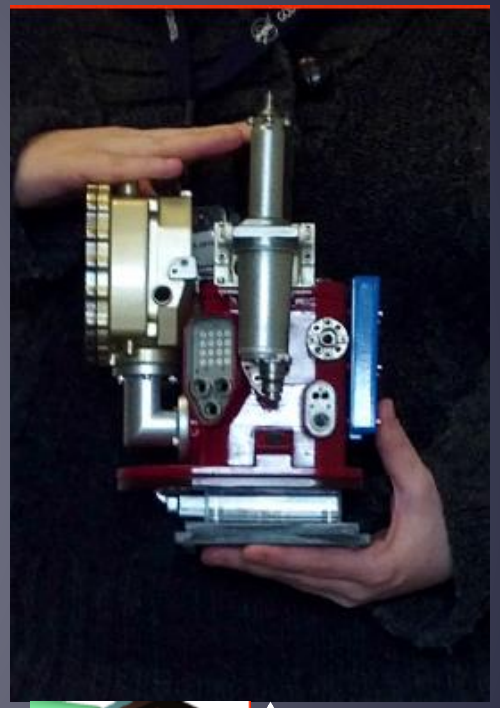
- Mars Organic Molecule Analyzer (MOMA) is an instrument suite on the ESA ExoMars 2018 Rover
  - **Mass Spectrometer (MS) – NASA/GSFC**
  - Sample Ovens – MPS
  - Gas Chromatograph (GC) – LISA and LATMOS
  - Laser Desorption (LD) - LZH
- Delivery Date for Flight MS: Spring 2016
- Scheduled Launch Date: May 2018
- D



The ExoMars rover. *Credit: ESA*

MOMA Hardware bioburden requirements

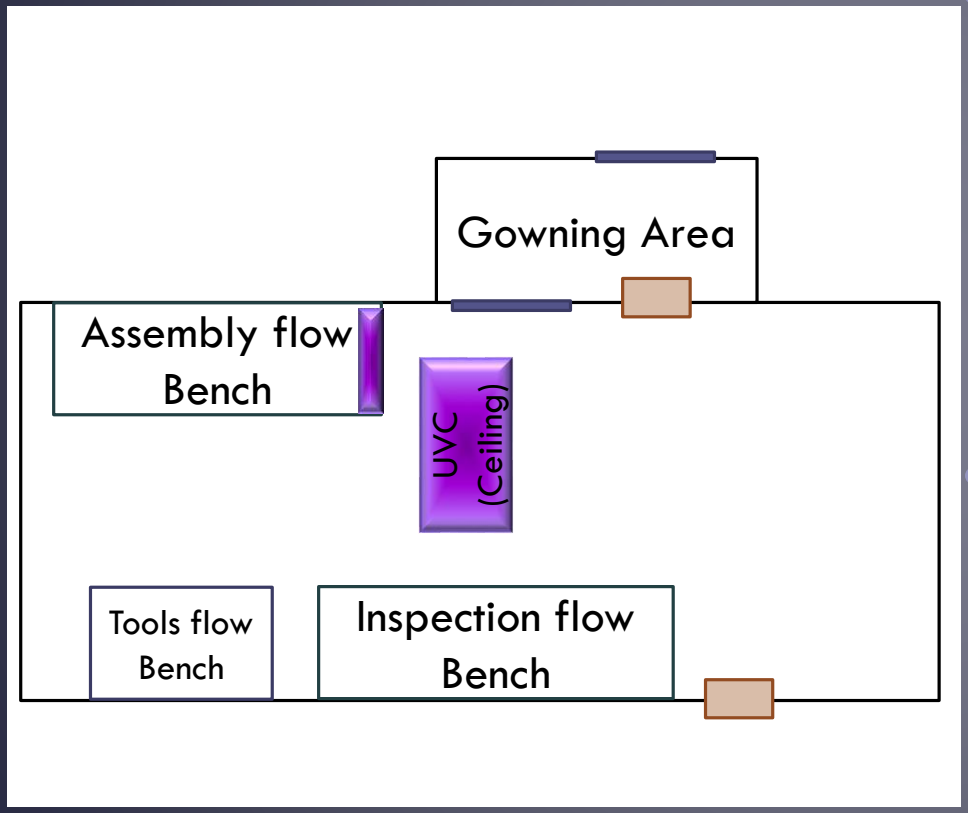
- Sample path (Ultra Clean Zone): **<0.03 spores/m^2**
  - Accessible area:
    - Base of MS
    - Internal surface of pseudo-Ultra Clean Zone (pUCZ)
  - Inaccessible area:
    - Internal surfaces of the Mass Spectrometer (MS)
    - Internal surfaces of Wide Range Pump (WRP)
    - Internal surfaces of Gas Processing System (GPS)
- Surfaces not in contact with sample path: **<1000 spores/m^2**
  - Exterior of MS, pUCZ, WRP, GPS,
  - Internal and external surfaces of Electronics boxes



Establishing clean working space and handling for MOMA-MS

- Three cleanrooms during build, integration, and testing
  - MOMA MS build:
    - Smallest cleanroom
    - Highest and continual bioburden control
  - Integration of MS and associated components, testing
    - Larger cleanroom, additional ULPA filter tent for sensitive integration steps
    - Bioburden control to be added as needed
  - Vacuum chamber and Mars environment testing
    - Custom vacuum chamber for environmental testing with clean tent
    - Bioburden control to be added as needed

MOMA MS build clean room

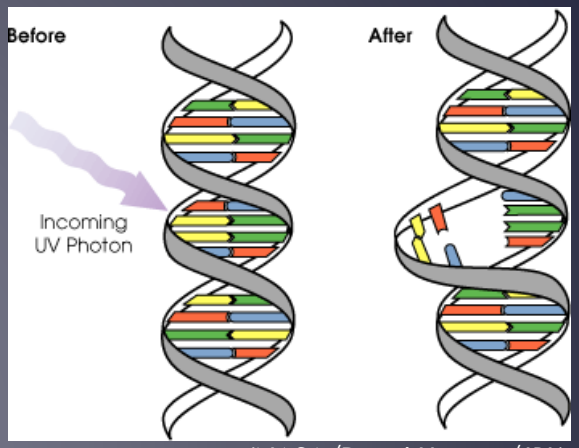


- Certified ISO class 7
- Maintains close to ISO 5

- Daily
  - Mop with weekly alternations between 70% IPA and 7.5% H_2O_2
  - Daily wipe of critical surfaces with sterile 70% IPA
- Twice a week:
  - Wipe horizontal surfaces with 100% IPA
  - Replace all garments
  - Run UVC lamps

Ultraviolet Light treatment of MOMA assembly cleanroom

- Ultraviolet-C (UV-C 100-290nm), 250-260nm is germicidal. Kills by crosslinking DNA, which prevents the organisms from replicating its DNA
- 22,000 $\mu Ws/cm^2$ is a sufficient energy dose to kill 99% of most common bacteria and bacterial spores on an exposed surface
- UV-C lamps (253nm) installed in cleanroom ceiling and on wall of assembly clean bench
- UV-C intensity at the floor of the cleanroom was measured at 30 $\mu W/cm^2$, 15 min exposure to reach 22,000 $\mu Ws/cm^2$

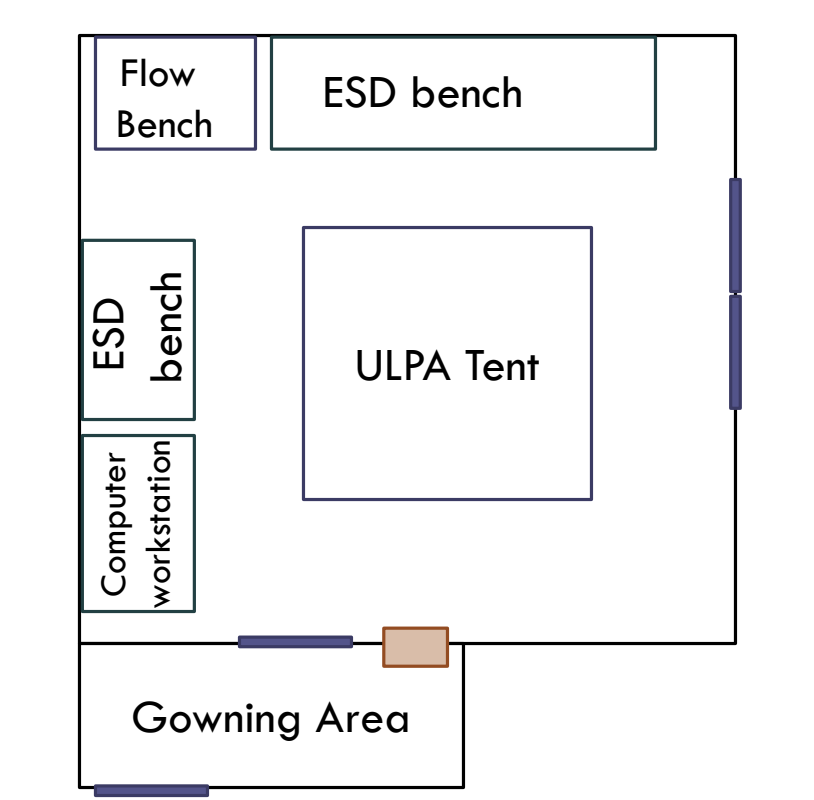


(NASA/David Herring/JPL)

Biocidal mopping

- Cleanroom mopped daily (M-F) with either 70% IPA or 7.5% H_2O_2
  - Alternate between IPA and H_2O_2 weekly
- Different biocidal mechanisms to prevent selecting for resistant organisms
  - 70% IPA denatures proteins
    - (70% IPA is a more effective biocide than 100% IPA)
  - 7.5% H_2O_2 disinfects by oxygen radical damage to DNA and proteins

MOMA-MS Integration &Testing clean room



- Certified ISO class 7
- Maintains close to ISO 6

- Daily:
  - Vacuum
- Twice a week
  - Mop with 5% IPA
  - Wipe horizontal surfaces with 100% IPA
  - Replace all garments
- Bioburden control to be instituted as necessary
  - During sample path exposure post DHMR

MOMA Planetary Protection Lab

- New capacity at Goddard Space Flight Center to support MOMA-MS
  - On-site planetary protection assay support allows closer monitoring and faster results
- Lab Development
  - Initial lab setup from July 2014, first MOMA-MS hardware samples processed November 2014
  - "*All operations involving the manipulation of sterile items and sample processing shall be performed in laminar flow environments meeting at least Class 100 air cleanliness requirements*" -NASA-HDBK-6022
  - Biological safety Cabinet class II type A2
    - Meets ISO class 5/ class 100 conditions
    - Provides both product and personnel protection
    - 70% air recirculation, HEPA filtration for cabinet and exhaust

(http://www.labconco.com/)

MOMA Planetary Protection Lab

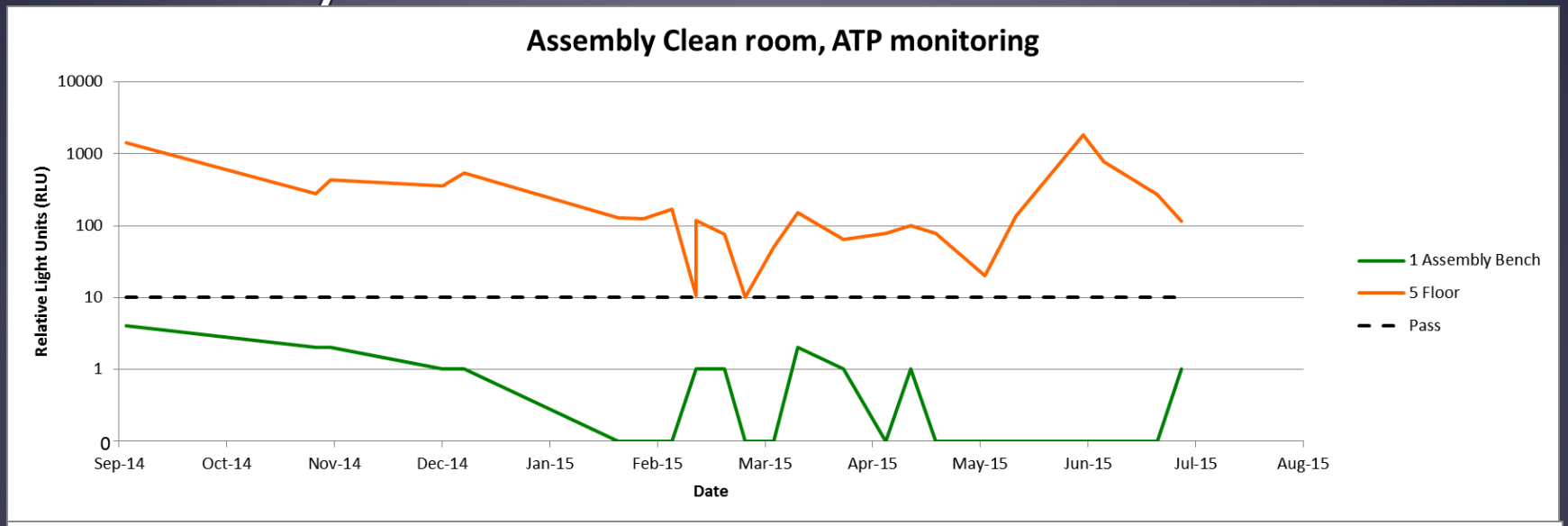
- Planetary Protection functionalities:
  - Rapid assay (ATP) (5min)
  - Testing airborne microbes (4 days)
  - Standard swab assay (4 days)
  - Autoclave sterility verification (2 days)
- Short term capacity expansion
  - Wipe assay for larger surface areas
  - DHMR (Dry Heat Microbial Reduction) verification
  - Biodiversity testing
- Long term plans to collaborate and share space with Astrobiology

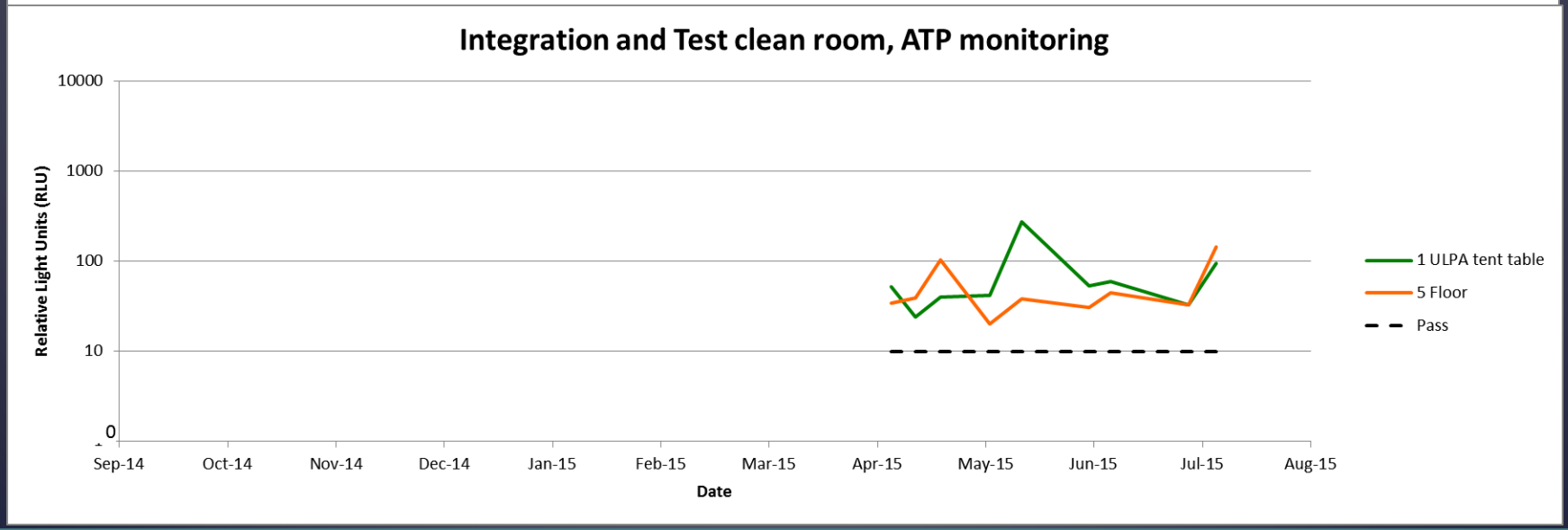
ATP rapid Bioburden Assessment


Everystockphoto.com/Clearly Ambiguous

- Pre-wet swab is used to sample a surface, swished in the reactant buffer.
  - ATP is the energy carrying molecule in all cell types
  - ATP in the sample will react with the luciferase and luciferin in the buffer and produce light
  - Less than 5 minutes to sample
- Pre-wet swab contains *Chlorhexidine digluconate*
  - Not to be used on sensitive hardware
  - Removable by 70% IPA wiping

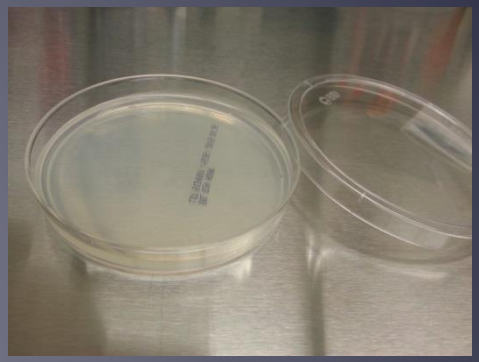
Critical work surfaces in assembly cleanroom have very low bioburden





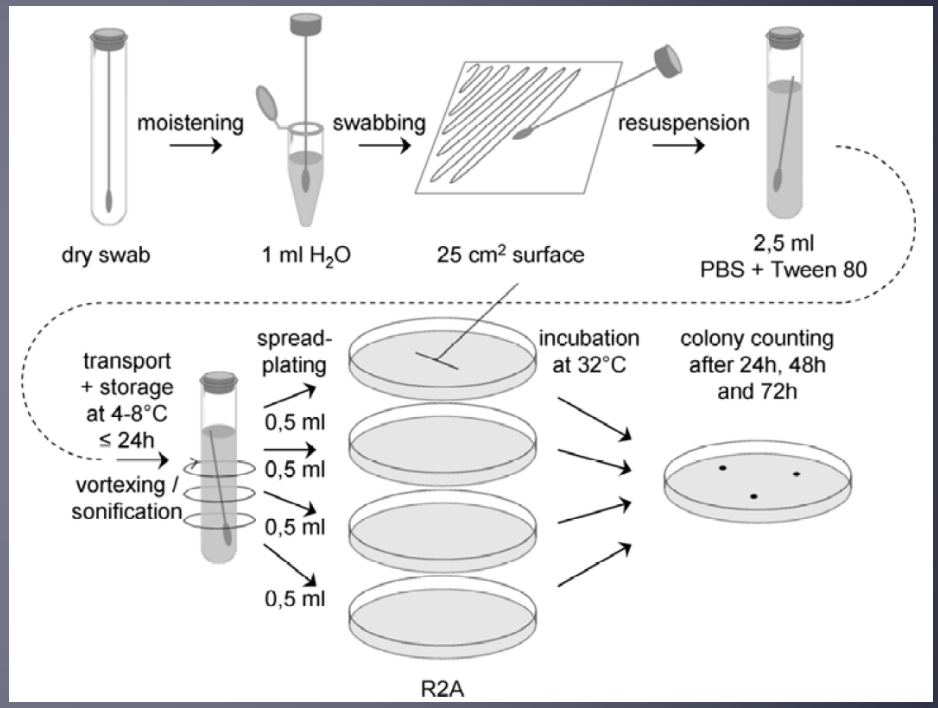
Airborne microbial monitoring

- Passive monitoring: Allowing airborne microbes to settle onto a plate surface.
  - Requires review in NASA cleanrooms because of high volatile content of plates
- Active monitoring: pulling air through a filter which is later transferred to a plate.
  - Used in MOMA cleanrooms:
    - **No growth yet seen in weekly cleanroom samples**
  - Will be used to monitor immediate environment during highly sensitive activities



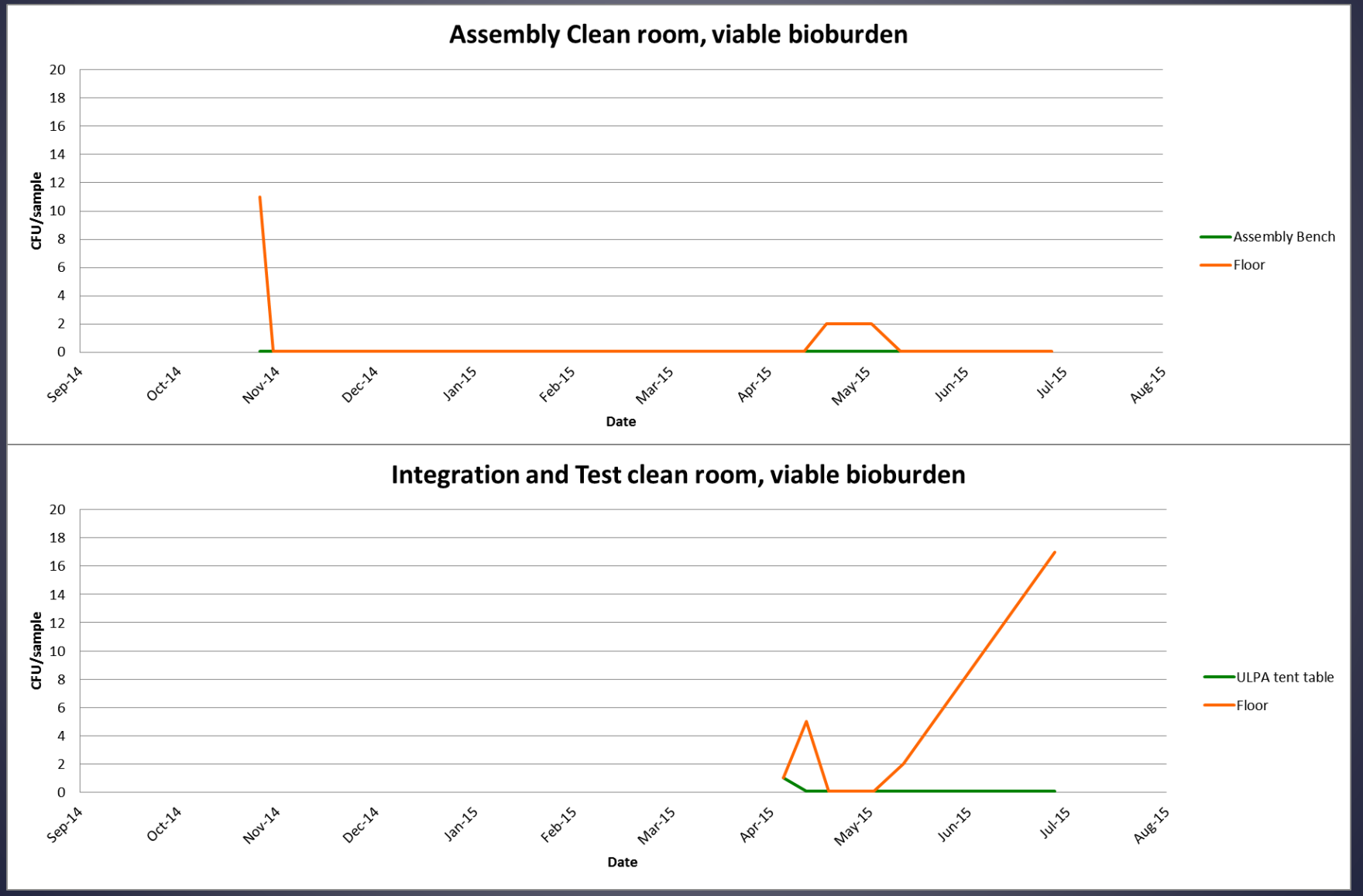
Facility bioburden monitoring

- Bioburden swabs in assembly and I&T cleanrooms
  - General viable microbe screen (not spore specific)
  - Swab a 25cm^2 area on work surface with a damp flocked nylon swab
  - Sample transported in 2.5ml sterile water
  - Processed by ESA protocol: ECSS-Q-ST-70-55C



(ECSS-Q-ST-70-55C, D.2.1)

Consistently low viable microbe counts

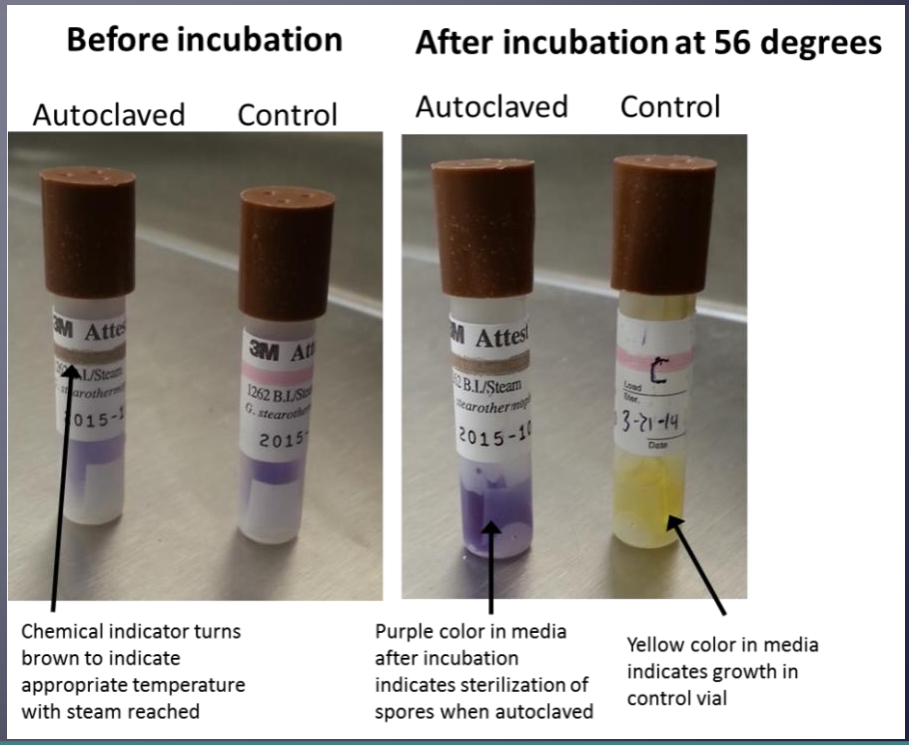


Post DHMR handling and cleanroom maintenance

- All sample path bioburden testing occurs prior to final access before DHMR
  - Post DHMR testing risks recontamination of the surface, and bioburden will be below limit of detection
- Any access to sample path post DHMR must occur in an aseptic ISO5 environment
  - Sterile garments, gloves, and tools required
  - Workspace tested for bioburden before work, actively monitored with air bioburden sampler

Post DHMR biological contamination prevention - Tools

- Tool management
  - After precision cleaning and white light inspection, compatible tools will be sterilized by autoclave
    - Autoclave sterilizes with heat + pressure + steam. 20 min 121°C
  - Biological indicator included in autoclave process to ensure sterilization
  - Tools that are not compatible with sterilization will not be used in direct contact with sample path surfaces post DHMR



Tool/Part Sterility

- Must only be exposed to ISO-5 or cleaner aseptic conditions
- Must be handled wearing sterile gowning
- Only wiped with sterile wipes
- Must only be set on sterile surfaces, sterile fields
- Must be opened by an assistant who is not handling sterile items
- Packages of foil will be sterilized for sterile fields (working surfaces)
- Sterile fields are single use and only for the continuous working session

Biological contamination prevention - Personnel

- Personnel management
  - One day Planetary Protection/ aseptic processing training for all personnel working directly with flight hardware
  - Single use sterile cleanroom coveralls, hood, and gloves
  - Two person system to manage sterile tools (pass sterile tool into workspace as needed)
- Sample path work only in an aseptic ISO 5 environment that has been verified by bioassay
- No tools that have not been sterilized in contact with Sample path

Summary

- MOMA-MS planetary protection requirements require the establishment of aseptic work spaces during assembly, integration, and testing
  - 2 cleanrooms will be used at GSFC
  - Assembly cleanroom is currently maintained with additional bioburden control steps, and maintains a very low level of biological contamination
  - Integration and Testing clean room has not had additional bioburden control steps instituted, and has a higher level of biological contamination
- New laboratory capacity at GSFC to process planetary protection samples
- After DHMR exposures of sample path will be limited
  - Opened only in a monitored aseptic work space with
  - Sterile garments, sterile tools

Acknowledgements

- MOMA-MS contamination Control and Planetary Protection team:
  - Radford Perry
  - Dr. John Canham
  - Lisa Crisp
  - Interns: Lynorra Grant (2014) and David Berlin (2015)
- MOMA-MS team and GSFC Code 699 (Planetary Environment Laboratory)
- GSFC Code 546 (Contamination and Coatings Engineering)
- GSFC Code 541 (Materials Engineering)